Determination of Young’s modulus of samples of arbitrary thickness using force microscopy

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Abstract

We present simple expressions for load required to indent a layer of arbitrary thickness with a conical, paraboloidal and cylindrical punch. A rigid substrate underneath the sample leads to an increase of load required for indentation. This effect has to be corrected for to prevent overestimation of Young’s modulus from force - distance curves, recorded with the Atomic Force Microscope (AFM). The problems of the frictionless contact of an axisymmetric punch and an isotropic, linear-elastic layer are reducible to Fredholm integral equations of the second kind. We solved them numerically and used the Remez algorithm to obtain piecewise polynomial approximations of the load indentation relation for samples that are either in frictionless contact with the rigid substrate or bonded to it. Their relative error due to approximation is negligible and uniformly spread. Combining the numerical approximations with asymptotic solutions for very thin layers, we obtained equations appropriate for samples of arbitrary thickness. They were implemented in a new version of AtomicJ, our free, open source application for analysis of AFM recordings.

Keywords: Contact problem, Elastic layer, Punch, Indentation, AFM

1. Introduction

The Atomic Force Microscope (AFM) allows for investigation of mechanical properties of materials through measurements of forces between the sample and a tip. The tip is mounted at the free end of a cantilever, which acts as a force sensor. The most widely used method to examine mechanical properties of a sample with an AFM is to measure the force acting on the cantilever when the tip is pressed into the sample. The recorded relation between the position $z$ of the fixed base of the cantilever and the force $P$ acting on the tip is known as a force - distance curve. The presence of a rigid substrate underneath the sample

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leads to an increase of load required for indentation. If this effect is not taken into account during data analysis, the obtained values of Young’s modulus are overestimated [1], [2]. This makes it difficult to separate the effects of topography from the true variability of the mechanical properties of the sample, e.g., in the case of animal cells forming thin cytoplasmic protrusions [3], [4].

To extract mechanical properties of the sample from a force-distance curve, it is necessary to assume a theoretical model of its contact with the tip, treated as a rigid punch. The theory of frictionless contact between a punch and a layer supported by a substrate is among the most studied topics in contact mechanics. The problem of an axisymmetric punch pressed into a non-bonded, isotropic layer, resting on a rigid half-space, was studied by Lebedev and Ufliand [5], who reduced it to a Fredholm integral equation of the second kind. Similar studies have been carried out for a layer bonded to the rigid substrate [6] and a layer bonded to a compliant, isotropic half-space [7], [11]. The more general problem of a stratified layer, supported by a half-space, has also received much attention [12]–[16].

The magnitude of the effect of the rigid substrate depends on the ratio \( \tau \) of the contact radius \( a \) (the radius of the area of direct contact between the punch and the sample, see Fig. 1) and the sample thickness \( h \). Methods for derivation of asymptotic formulas for load \( P \) and indentation \( \delta \) have been presented in [17], [18], [19] (for \( \tau \ll 1 \)) and in [20] (for \( \tau \gg 1 \)). These formulas have a parametric form - both load and the indentation depth are expressed as functions of a parameter which is not directly measured, so that numerical calculations are necessary to find the load - indentation relation. Formulas for load expressed as an explicit function of \( \delta \) are more convenient. Approximations of \( P(\delta) \), based on the assumption of small \( \tau \), have been derived in [21], [22] and [23] for a paraboloidal punch, and in [22], [4], [24] and [25] for a conical punch. Analogous approximations of \( P(\delta) \) appropriate for large values of \( \tau \) have been proposed in [26] for conical and paraboloidal indentation.

During development of AtomicJ [27], our open source application for analysis of AFM recordings, we became aware of the need to develop polynomial approximations of load - indentation functions for samples of arbitrary thickness, with a small and uniformly spread error. Force - distance curves are used to assess elastic properties of diverse samples, whose thickness varies from a few nanometres in the case of lipid layers to millimetres. Thus, approximations appropriate for implementation in a publicly available software should ensure accurate calculations for a wide range of sample thickness, preferably for all non-negative values of the parameter \( \tau \).

In this paper, we present simple, piecewise polynomial approximations of \( P(\delta) \) for frictionless contact of a conical, paraboloidal or a cylindrical punch with an isotropic, linear elastic layer resting on a rigid substrate. Their error is low and uniform. The approximates have been implemented in AtomicJ. We also compared the numerical solutions with formulas known from the literature, including those frequently used to analyse force - distance curves.
Figure 1: Indentation of a layer of thickness $h$, resting on a rigid substrate, with an axisymmetric punch. The depth of indentation is denoted as $\delta$, contact radius as $a$ and load as $P$. The $z$ axis is directed towards the substrate.

2. Contact problem for an elastic layer

We will consider a frictionless, normal contact between a linear-elastic layer of thickness $h$ and a rigid, axisymmetric punch (fig. 1). The punch profile is described by a smooth function $f(\varrho)$, satisfying $f(0) = 0$. The upper surface ($z = 0$) of the layer is loaded by the punch. The lower surface ($z = h$) is supported by a rigid substrate of infinite thickness. Due to the axial symmetry, the region of direct contact between the layer and the punch is a disk of radius $a$. Circumferential displacements vanish everywhere. The boundary conditions on the upper ($z = 0$) surface of the layer are

$$
\begin{align*}
  u_z(\varrho, 0) &= \delta - f(\varrho) & & 0 \leq \varrho \leq 1 \\
  \sigma_z(\varrho, 0) &= 0 & & \varrho > 1 \\
  \tau_{rz}(\varrho, 0) &= 0
\end{align*}
$$

where $\varrho = \frac{r}{a}$ is the normalized radial coordinate, $u_z(\varrho, z)$ is normal displacement, $\sigma_z(\varrho, z)$ is normal stress, $\tau_{rz}(\varrho, z)$ is shear stress, $\delta$ is the depth of indentation. The conditions at the lower surface depend on the type of the contact with the substrate. If the contact with the substrate is frictionless and the displacement of points on the lower sample surface is restricted only in the normal direction, then

$$
\begin{align*}
  u_z(\varrho, h) &= 0 \\
  \tau_{rz}(\varrho, h) &= 0
\end{align*}
$$

If the sample is bonded to the substrate, the points in contact with the substrate cannot move in the radial or normal direction

$$
\begin{align*}
  u_z(\varrho, h) &= 0 \\
  u_r(\varrho, h) &= 0
\end{align*}
$$

The contact problems specified by the above conditions resemble the classical Hertz problem for a punch of a circular planform, except for the finite thickness of the sample. The limitations of the Hertz-type contact models and the alternative approaches to contact modelling are discussed in [28]. The problem of
a non-bonded elastic layer, resting on a rigid substrate (conditions (1) – (5)), was studied by Lebedev and Ufliand [5]. The problem of a bonded layer, described by (1) – (3) and (6) – (7), was studied by Pupyrev and Ufliand [6]. Both problems were reduced to a Fredholm integral equation of the second kind

\[
\chi(x; \tau) = \chi^\infty(x) + \frac{1}{\pi} \int_0^1 \chi(t; \tau) K(x, t; \tau) \, dt
\]  

(8)

We will refer to (8) as the Lebedev-Ufliand equation. Its solution \( \chi(x) \) can be used to calculate parameters describing the tip-sample contact, including load and contact radius. Indentation depth and the punch profile enter the equation through its free term \( \chi^\infty(x) \)

\[
\chi^\infty(x) = \frac{2}{\pi} \left[ \delta - x \int_0^x \frac{df(z)}{\sqrt{x^2 - z^2}} \, dz \right]
\]

(9)

The kernel \( K \) of (8) can be expressed as

\[
K(x, t; \tau) = \Omega(t + x; \tau) + \Omega(t - x; \tau)
\]

(10)

where \( \Omega(t + x; \tau) \) is a cosine transform of the weight function \( \omega(p) \)

\[
\Omega(y; \tau) = \int_0^\infty \omega(p; \tau) \cos(py) \, dp
\]

(11)

The type of contact between the layer and the substrate determines the form of \( \omega(p) \). For a non-bonded layer, \( \omega(p) \) depends on one parameter \( \tau = \frac{a}{h} \). It can be expressed as [5]

\[
\omega(p; \tau) = 1 - \frac{2 \sinh^2\left(\frac{p}{\tau}\right)}{2p + \sinh\left(\frac{2p}{\tau}\right)}
\]

(12)

For a bonded layer, \( \omega(p) \) depends on two parameters, \( \tau \) and the Poisson ratio \( \nu \) [6]

\[
\omega_b(p; \tau, \nu) = \frac{(3 - 4\nu)^2 + (1 + 2\nu)^2 + 2(3 - 4\nu)e^{-2\frac{p}{\tau}}}{(3 - 4\nu)e^{2\frac{p}{\tau}} + (3 - 4\nu)^2 + (1 + 4\nu) + (3 - 4\nu)e^{-2\frac{p}{\tau}}}
\]

(13)

\( \Omega(y; \tau) \) approaches zero when \( \tau \to \infty \). In this limiting case, the solution \( \chi \) becomes equal to \( \chi^\infty \).

The solution \( \chi \) of (8) can be used to calculate normal stress at the upper surface \( \sigma_z(q, 0) \), according to the equation

\[
\sigma_z(q, 0) = -\frac{E}{2a(1 - \nu^2)} \left( \frac{\chi(1)}{\sqrt{1 - q^2}} - \int_q^1 \frac{d\chi(t)}{\sqrt{t^2 - q^2}} \, dt \right) \quad q \leq 1
\]

(14)

The load \( P \) can be calculated by integration of \( \sigma_z(q, 0) \) within the area of contact between the punch and the layer. The final expression is

\[
P = \pi a E \int_0^1 \chi(t) \, dt
\]

(15)
where \( E \) is the Young’s modulus of the layer.

The boundary conditions for the problem of a punch indenting a layer do not determine the contact radius \( a \), i.e. the radius of the area of direct contact between the punch and the layer. If forces of adhesion are absent, an additional condition of finite stress at the edge of the contact area must be introduced. In accordance with \([14]\), it leads to the criterion \([5]\)

\[
\chi(1; \tau) = 0
\]

More general criteria for the contact radius are provided by the Johnson-Kendall-Roberts (JKR) model of adhesive contact, which can be extended in a natural way to the contact between a punch and a thin layer \([29, 30] \).

\textit{Punch profiles.} In this work, we will consider power-law-shaped punches, with profiles described by

\[
f(\varrho) = B a^\eta \varrho^\eta
\]

where \( \eta \geq 1 \). Important special cases of such punches are a cone with the half angle \( \theta \) (\( \eta = 1, B = \frac{1}{\tan(\theta)} \))

\[
f(\varrho) = \frac{1}{\tan(\theta)} a \varrho
\]

and a paraboloid with the radius of curvature \( R \) (\( \eta = 2, B = \frac{1}{2R} \))

\[
f(\varrho) = \frac{1}{2R} a^2 \varrho^2
\]

A cylindrical punch of the radius \( a < 1 \) can be considered as a limiting case of a power-law-shaped punch, with \( \eta \to \infty \).

The exact profile of a spherical punch of radius \( R \) is

\[
f(\varrho) = R - \sqrt{R^2 - a^2 \varrho^2}
\]

The paraboloidal profile \([19]\) is the first non-zero term in the Taylor expansion of the spherical profile \([20]\) at the punch apex \( \varrho = 0 \). For this reason, the load - indentation relations for a paraboloid are often described as equations for a sphere. However, the case of a punch with the exact spherical profile has been also analysed in the literature (e.g. \([18, 31]\)), so here, we will refer to the punches described by \([19]\) as paraboloidal.

3. \textbf{Formal expressions for load and contact radius}

The solution of a Fredholm integral equation of the second kind can be formally represented using the resolvent \( H \), which is independent of the free term of the equation. The resolvent corresponding to \([8]\) depends on the same
parameters as the kernel $K$. Thus, $H$ depends on $\tau$ and, in general, on $\nu$, which we will reflect in our notation by writing $H(x,t;\tau,\nu)$. The solution $\chi$ is

$$\chi(x;\tau) = \chi^\infty(x) + \frac{1}{\pi} \int_0^1 H(x,t;\tau,\nu)\chi^\infty(t) \, dt \quad (21)$$

To express load and contact radius in terms of the resolvent $H$, we will introduce the notation

$$\Upsilon^{(\eta)}(\tau;\nu) = \frac{1}{\pi} \int_0^1 H(1,t;\tau,\nu)t^\eta \, dt \quad (22)$$

$$\Xi^{(\eta)}(\tau;\nu) = 1 + \frac{\eta}{\pi} \int_0^1 \int_0^1 H(x,t;\tau,\nu)t^\eta \, dt \, dx \quad (23)$$

The variation of $\Upsilon^{(\eta)}$ and $\Xi^{(\eta)}$ with $\tau$ for a non-bonded layer is shown in fig. 2.

**Equations for contact radius.** In accordance with (16) and (21), the equilibrium value of the contact radius $a$ can be calculated from the equation

$$0 = \chi^\infty(1;a) + \frac{1}{\pi} \int_0^1 H(1,t;\tau,\nu)\chi^\infty(t;a) \, dt \quad (24)$$

Combining (9) and (24), we obtain an equation connecting the equilibrium value of contact radius with indentation depth. For a punch of power-law-shaped profile (17), we arrive at

$$\kappa = \sqrt{\frac{\pi \tau \eta}{2 \Gamma\left(\frac{\eta}{2}\right)}} \left[ 1 + \frac{\Upsilon^{(\eta)}(\tau,\nu)}{1 + \Upsilon^{(0)}(\tau,\nu)} \right]$$

(25)
Figure 3: The parameter $\kappa$ (eq. 26) as a function of the normalized contact radius $\tau = \frac{a}{h}$, for a non-bonded layer. The calculations were carried out for a non-adhesive contact with a power-law-shaped punches of $\eta$ equal to 1 (cone, dotted line), 2 (paraboloid, dashed line) and 3 (solid line).

$\kappa = \frac{\delta}{\eta Bh^n}$  \hspace{1cm} (26)

The dimension of $B$ is length to the power of $1 - \eta$, so that $\kappa$ is a dimensionless parameter. For a cone, $\kappa$ is equal to $\frac{\delta}{h} \tan(\theta)$, for a paraboloid $\kappa$ equals $R \delta h^2$. The dependence of $\tau$ from $\kappa$ is shown in Fig. 3.

Equations for load. The solution $\chi(x)$ of the Lebedev-Ufliand equation for a thin sample can be expressed by combining the formula (21) for $\chi$ in terms of the resolvent and the solution (9) for the infinitely thick sample. For a power-law-shaped punch

$$\chi(x) = \frac{2}{\pi} \left[ \delta - \frac{\sqrt{\pi \eta Ba^n \Gamma(\frac{n}{2}) x^n}}{2 \Gamma(\frac{1+n}{2})} \right] + \frac{1}{\pi} \int_0^1 H(x, t; \tau, \nu) \frac{2}{\pi} \left[ \delta - \frac{\sqrt{\pi \eta Ba^n \Gamma(\frac{n}{2}) t^n}}{2 \Gamma(\frac{1+n}{2})} \right] dt$$  \hspace{1cm} (27)

To find the equation for load $P$, we substitute (27) to the general equation (15) describing the relation between $P$ and $\chi$. After simplification

$$P = \frac{aE}{1 - \nu^2} \left[ 2 \delta \left( 1 + \Xi^{(0)}(\tau, \nu) \right) - \left( 1 + \Xi^{(n)}(\tau, \nu) \right) \frac{\sqrt{\pi \eta Ba^n \Gamma(1 + \frac{n}{2})}}{\Gamma(\frac{1+n}{2})} \right]$$  \hspace{1cm} (28)

If $\tau = 0$, then $\Xi^{(n)} = 0$ for any $\eta$. In such a case, (28) becomes identical with the equation for load necessary to indent an infinitely thick sample.

$$P = \frac{aE}{1 - \nu^2} \left( 2 \delta \frac{\sqrt{\pi a^n B \Gamma(1 + \frac{n}{2})}}{\Gamma(\frac{1+n}{2})} \right)$$  \hspace{1cm} (29)

If adhesion forces are absent, the equilibrium value of the contact radius is

$$a = \sqrt[\frac{n}{2}]{\frac{2 \delta \Gamma(\frac{1+n}{2})}{\sqrt{\pi \eta B \Gamma(\frac{n}{2})}}}$$  \hspace{1cm} (30)

Substituting (30) to (29), we obtain

$$P = \frac{2\eta E\delta^{\frac{2+n}{2}}}{(1 - \nu^2)(1 - \eta)} \sqrt[\frac{n}{2}]{\frac{2 \Gamma(\frac{1+n}{2})}{\sqrt{\pi \eta B \Gamma(\frac{n}{2})}}}$$  \hspace{1cm} (31)
This equation was derived by Shtaerman \[32\] (for even η) and Galin \[33\] (for any real η ≥ 1).

A cone with the half-angle θ is a power-law-shaped punch with η = 1 and B = 1 / tan(θ). Load can be expressed as

\[
P = \frac{Ea \left[ 2\delta(1 + \Xi^{(0)}) - \frac{\delta}{\nu}\cot(\theta)(1 + \Xi^{(1)}) \right]}{1 - \nu^2}
\] (32)

When adhesion forces are absent, the contact radius can be calculated from \(a = \frac{\delta}{\pi}\tan(\theta)\). In such a case, \[32\] assumes the form \[34, 35\]

\[
P = \frac{2E\tan(\theta)\delta}{\pi(1 - \nu^2)}
\] (33)

This equation is often referred to as Sneddon’s model.

A paraboloid of radius R is a power-law-shaped punch with η = 2 i B = 1 / 2R. Thus, load can be expressed as

\[
P = \frac{2Ea \left[ 3R\delta(1 + \Xi^{(0)}) - a^2(1 + \Xi^{(2)}) \right]}{3(1 - \nu^2)R}
\] (34)

If the contact is non-adhesive, then \(a = \sqrt{R\delta}\). In this case, load can be calculated from Hertz’s equation \[36\]

\[
P = \frac{E}{1 - \nu^2} \frac{4}{3} \sqrt{R\delta}\frac{a^2}{R}
\] (35)

For a cylindrical punch, load can be found as

\[
P = \frac{aE\delta}{1 - \nu^2} \left[ 2 \left( 1 + \Xi^{(0)} \right) \right]
\] (36)

The relationship between load and the depth of indentation by a cylindrical punch is linear, regardless of sample thickness. Influence of the rigid substrate on load does not depend on indentation depth.

In \[28 - 36\], the effect of the rigid substrate is captured by the parameters \(\Xi^{(n)}\). For fixed η, they are functions of τ (non-bonded) or τ and ν (bonded layer). They are not directly dependent on the punch size. Parameters \(\Xi^{(0)}\) and \(\Xi^{(1)}\) in the equation \[32\] for load in conical indentation are independent of the cone half-angle. Likewise, \(\Xi^{(0)}\) and \(\Xi^{(2)}\) in the equation \[34\] are independent of the radius of the paraboloid.

4. Approximations of load for conical, paraboloidal and cylindrical tips in the absence of adhesion forces

To approximate load required to indent a layer with a conical or paraboloidal tip, we will use a dimensionless form of \[28\]

\[
\Lambda = \tau \left[ 2 \left( 1 + \Xi^{(0)} \right) - \left( 1 + \Xi^{(n)} \right) \frac{\sqrt{\pi}\tau^\eta \Gamma(1 + \frac{\eta}{2})}{\eta \Gamma(\frac{1 + \eta}{2})} \right]
\] (37)
where $\Lambda$ is dimensionless load

$$\Lambda = \frac{P(1 - \nu^2)}{\eta B E h^{\nu+1}}$$

(38)

In the special case of conical indentation $\Lambda$ is equal to $\frac{P \tan \theta (1 - \nu^2)}{E h^2}$. It can be calculated from

$$\Lambda = \tau \left[ 2 \kappa (1 + \Xi^{(0)}) - \frac{\pi}{2} \tau (1 + \Xi^{(1)}) \right]$$

(39)

For paraboloidal indentation, $\Lambda = \frac{PR(1 - \nu^2)}{E h^3}$. It can be calculated as

$$\Lambda = \tau \left[ 2 \kappa (1 + \Xi^{(0)}) - \frac{2}{3} \tau^2 (1 + \Xi^{(2)}) \right]$$

(40)

If adhesion forces are absent, the equilibrium value of $\tau$ is a function of $\kappa$ (non-bonded) or $\kappa$ and $\nu$ (bonded layer). Thus, dimensionless load can also be treated as a function of $\kappa$ and $\nu$. Cylindrical indentation requires a different definition of dimensionless load. We will define it as

$$\Lambda = \frac{P(1 - \nu^2)}{2a E \delta}$$

(41)

We approximated $\Lambda$ with low-degree polynomials $\hat{\Lambda}$. The goal of approximation was to keep the maximal relative error below $10^{-3}$. Such error is negligible compared to other sources of uncertainty, including the experimental measurement errors and simplifications inherent in the mathematical formulation of the contact problem through linear boundary conditions. In the first step, $\Lambda(\kappa)$ was expanded as a combination of Chebyshev polynomials of high order (20) using interpolation through Gauss - Chebyshev - Lobatto nodes $\kappa_i$. These approximations were used as proxies for calculation of less accurate, but simpler expressions. For a non-bonded and a bonded incompressible layer, the final approximants were calculated using the Remez algorithm, available in Mathematica (Wolfram Research, Illinois). For a bonded compressible layer, weighted least squares were used instead. For small $\kappa$, polynomials in $\kappa^1 \eta$ were used for approximation, as suggested by the asymptotic analysis [22]. For medium and large $\kappa$, the approximants are polynomials in $\kappa$.

To calculate reduced load in a particular node $\kappa_i$, we must first find the corresponding equilibrium value of contact radius. Based on the criterion [16], we searched with Newton’s algorithm for a root of $\chi(1; \tau)$, treated as a function of $\tau$. Each evaluation of $\chi(1; \tau)$ requires solving Lebedev - Uffliand equation [8], using the Nyström method [37] with a Gauss - Chebyshev - Radau quadrature that contains a node at 1. The number of nodes necessary to calculate contact radius increases with $\kappa$ (see Supporting Fig. S1). We used up to 1200 nodes, depending on $\kappa$. Once contact radius corresponding to $\kappa_i$ is known, reduced load can be calculated from (37) or (41). The parameters $\Xi^{(n)}$ were obtained from

$$\Xi^{(n)} = (1 + \eta) \int_0^1 [\chi(x) - x^n] \, dx$$

(42)
where \( \chi \) is the solution of
\[
\chi(x) = x^\eta + \frac{1}{\pi} \int_0^1 K(x,t)\chi(t)\,dt
\]  
\((43)\)
The formula \((42)\) can be derived by combining \((21)\) with \(\chi^\infty(x) = x^\eta\) and \((23)\). We used a Gauss-Legendre quadrature \(Q_N\) with \(N\) nodes to solve \((43)\). For each node \(x_i\), we discretized the equation \((43)\) by \(Q_N\) and then computed the values of \(\chi(x_j)\) \((j = 1, \ldots, N)\) using the Nyström method. The integral of \(\chi(x) - x^\eta\) with respect to \(x\) was subsequently calculated using the quadrature \(Q_N\) and the values \(\chi(x_j)\).

Solving the Lebedev-Ufliand equation requires multiple evaluations of the function \(\Omega\), given by \((11)\), which is a non-elementary cosine transform. To speed up calculations, we expanded \(\Omega\) into a series of the Christov functions, as discussed in the Appendix A.

**Conical tips.** Dimensionless load required to indent a very thin layer \((\tau \gg 1)\) with a conical punch can be calculated using asymptotic expressions, whose leading terms are \([26]\)
\[
\Lambda = \frac{\pi}{3} \kappa^3 \quad (44)
\]
\[
\Lambda = \frac{\pi}{80} \left(\frac{3}{2}\right)^5 \kappa^5 \quad (45)
\]
for a non-bonded and an incompressible bonded layer, respectively. For a cone, \(\kappa\) is equal to \(\frac{\delta \tan(\theta)}{h}\), in accordance with \((26)\). Numerical calculations indicate that the relative error of the equation \((44)\) for a non-bonded layer decreases fast with \(\kappa\), dropping below \(10^{-3}\) for \(\kappa > 4.1\) (fig. 4 b). Thus, the asymptotic formula \((44)\) was used when \(\kappa > 5\). For smaller \(\kappa\), the reduced load was approximated as
\[
\tilde{\Lambda}(\kappa) = \frac{2}{\pi} \kappa^2 (1 + 0.461\kappa + 0.346\kappa^2 + 0.0484\kappa^3) \quad \kappa \leq 1
\]  
\[
\tilde{\Lambda}(\kappa) = 0.0859 + 0.103\kappa - 0.0647\kappa^2 + 1.057\kappa^3 \quad 1 < \kappa \leq 5
\]  
\((46)\)  
\((47)\)

The relative error of the asymptotic expression \((45)\) for a bonded layer decreases much slower than the error of the corresponding expression \((44)\) for a non-bonded layer (fig. 5 b). It drops below \(10^{-3}\) only when \(\kappa > 88.9\). Thus, we calculated approximations in a much wider interval than in the case of non-bonded layer
\[
\tilde{\Lambda}(\kappa) = \frac{2}{\pi} \kappa^2 (1 + 0.715\kappa + 0.609\kappa^2 + 0.735\kappa^3) \quad \kappa \leq 0.9
\]  
\[
\tilde{\Lambda}(\kappa) = -0.265 + 1.225\kappa - 1.651\kappa^2 + 2.332\kappa^3 + \frac{\pi}{80} \left(\frac{3}{2}\right)^5 \kappa^5 \quad 0.9 < \kappa
\]  
\((48)\)  
\((49)\)

For \(\kappa > 89\), the equation \((45)\) can be used. The relative errors of the derived approximations for load required to indent a layer with a conical tip are shown in fig. 4 a, b (non-bonded) and 5 a, b (bonded layer).
Figure 4: Relative approximation error $\epsilon$ of formulae for force required for indentation of a non-bonded layer with a conical (a, b) or paraboloidal (c, d) punch. Calculations were performed for expressions derived under the assumption of small (a, c) or large (b, d) $\tau$. Red solid lines show the error of the piecewise approximations presented in this work, eq. (46) - (47) in a, b and eq. (52) - (53) in c, d. Dotted lines show the error made when the equations for infinitely thick samples are used (Sneddon’s model [35], [34], marked as I in a - b, Hertz’s model [36] marked as VI in c - d). The approximation II was published in [4], III in [25], IV in [22], V in [26], VII in [21], VIII in [19], IX in [22], X in [20], XI in [26] and [38].

Paraboloidal tips. When $\tau$ is large, dimensionless load for paraboloidal indentation can be approximated as [15], [26, 38, 39]

$$\Lambda = \pi z^2$$  \hspace{1cm} (50)

$$\Lambda = \frac{\pi}{2} z^3$$  \hspace{1cm} (51)

for a non-bonded and an incompressible bonded layer, respectively. For a paraboloid, $z$ is equal to $\frac{Rd}{h^2}$, in accordance with [20]. Our calculations for a non-bonded layer indicate that the relative error of (50) is less than $10^{-3}$ when $z > 17.1$. The interval of approximation was split into three regions. In
Figure 5: Relative approximation error $\epsilon$ of formulae for force required for indentation of a bonded layer with a conical (a, b) or paraboloidal (c, d) punch. Calculations were performed for expressions derived under the assumption of small (a, c) or large (b, d) $\tau$. Red solid lines show the error of the piecewise approximations presented in this work, eq. (48) - (49) in a, b and (54) - (55) in c, d. Dotted lines show the error made when the equations for infinitely thick samples are used (Sneddon’s model [35], [34] marked as I in a, b, Hertz’s model [36] as VII in c, d). The approximation II was published in [4], III in [23], IV in [22] and [25], V in [26], VI in [40], VIII in [21], IX in [19], X in [22], XI in [26] and [38], XII in [40].

the first two of them

$$\tilde{\Lambda}(\kappa) = \frac{4}{3} \kappa^2 \left( 1 + 0.722 \kappa^2 + 0.822 \kappa \right) \kappa \leq 0.5$$

$$\tilde{\Lambda}(\kappa) = -0.0633 + 0.260 \kappa^2 + \pi \kappa^2 \quad 0.5 < \kappa \leq 450$$

In the third region of $\kappa > 450$, the asymptotic formula (50) was used. In the case of a bonded layer, the relative error of the asymptotic expression for very thin layers (eq. 44) drops below $10^{-3}$ only when $\kappa > 5313$. The corresponding
piecewise approximations of $\Lambda$ are

\[
\Lambda(\kappa) = \frac{4}{3} \kappa^\frac{3}{2} \left( 1 + 1.105 \kappa^\frac{1}{2} + 1.607 \kappa + 1.602 \kappa^\frac{3}{2} \right) \quad \kappa \leq 0.4 \quad (54)
\]

\[
\Lambda(\kappa) = 0.616 - 3.114 \kappa^\frac{1}{2} + 6.693 \kappa - 7.170 \kappa^\frac{3}{2} + 8.228 \kappa^2 + \frac{\pi}{2} \kappa^3 \quad 0.4 < \kappa \quad (55)
\]

We calculated the relative error of approximation (54) - (55) for $\kappa \leq 20000$. In this range, the error is negligible. The errors of the proposed approximations for paraboloidal tips are shown in fig. 4 c, d (non-bonded) and fig. 5 c, d (bonded layer).

**Cylindrical tips.** The leading terms of the asymptotic expansion of the dimensionless load $\Lambda$ required for cylindrical indentation of a very thin layer ($\tau \gg 1$) are [41]

\[
\Lambda = \frac{\pi}{2} \tau 
\]

\[
\Lambda = \frac{3\pi}{64} \tau^3 
\]

for a non-bonded and an incompressible bonded layer, respectively. We calculated approximations of $\Lambda$ separately in two intervals. For a non-bonded layer, we obtained

\[
\Lambda(\tau) = 1 + 0.725 \tau + 0.697 \tau^2 - 0.259 \tau^3 + 0.0347 \tau^4 \quad \tau \leq 1.8 \quad (58)
\]

\[
\Lambda(\tau) = 0.602 + 1.562 \tau + 0.000371 \tau^2 \quad 1.8 < \tau \leq 22 \quad (59)
\]

For $\tau > 22$, we used the asymptotic expansion derived in [15], truncated to the first two terms

\[
\Lambda = \frac{\pi}{2} \tau + 0.589
\]

(60)

For a bonded layer, the piecewise approximations take the form of

\[
\Lambda(\tau) = 1 + 1.101 \tau + 1.518 \tau^2 + 0.0207 \tau^3 \quad \tau \leq 0.8
\]

\[
\Lambda(\tau) = 0.713 + 1.893 \tau + 0.869 \tau^2 + 0.146 \tau^3 \quad 0.8 < \tau \leq 10
\]

(61)

(62)

If $\tau > 10$, then $\Lambda$ can be calculated with negligible error, using the asymptotic expansion proposed in [39], truncated to

\[
\Lambda = \frac{1}{4\beta^3} \left( 1 + 1.924\beta + 1.924\beta^2 - 2.520\beta^3 \right)
\]

(63)

where $\beta = \frac{1}{1 + \left(\frac{\tau}{16}\right)^3 \tau}$. The relative error of piecewise approximations of reduced load $\Lambda$ for a non-bonded and a bonded layer is shown in fig. 3 a, b.

The equations for conical, paraboloidal and cylindrical tips presented above do not describe indentation of a bonded, compressible layer. In such a case,
Figure 6: Relative error $\epsilon$ of piecewise approximations of dimensionless load $\Lambda$ for a cylindrical punch, indenting a non-bonded (a) or bonded (b) layer. Dashed line marks the errors equal to $10^{-3}$.

dimensionless load $\Lambda$ is also a function of Poisson’s ratio of the sample $\nu$. Numerical results indicate that the influence of $\nu$ on $\Lambda$ grows with $\kappa$. In particular, when the layer is very thin and nearly incompressible (i.e. with $\nu$ close to 0.5), small increments of $\nu$ leads to a large increase of $\Lambda$ (fig. 7). To approximate the relationship between $\Lambda$ and $\kappa$ with the relative error below $10^{-3}$, we had to split the domain $\kappa - \nu$ into multiple regions. As they are numerous, these equations are not included in this paper, but they can be found in the publicly available source code of AtomicJ.

5. Discussion

In this paper, we presented low-degree piecewise polynomial approximations of load required to indent a linear-elastic, isotropic layer with a conical, paraboloidal or cylindrical punch. The equations have been obtained under the assumption that the contact between the punch and the layer is frictionless, while the layer can either freely slip on the rigid substrate (the non-bonded case) or it is bonded to the substrate. Through combination of numerical techniques and known asymptotic formulas for very thin layers, we obtained equations that describe layers of arbitrary thickness. Comparison of the approximate formulae with our numerical solutions of the Lebedev - Ufliand integral equation [5] indicates that their relative error is smaller than $10^{-3}$. Thus, it is negligible compared to other contributions to the error of the values of Young’s modulus calculated from force - distance curves, for example uncertainty of cantilever’s spring constant, inhomogeneity of the sample, deviations from the assumptions of the linear elasticity or the effect of lateral displacements of the sample material, which are not accounted for by the hertzian conditions (1) - (3). The equations presented in this paper have been implemented in the new version of AtomicJ, freely available through the Source Forge platform. AtomicJ allows for concurrent processing of multiple force distance curves and force - volume recordings. For analysis of force curves recorded on a thin sample, the thickness can be automatically read from a user-specified topographical image.
Figure 7: The dependence of the dimensionless load $\Lambda$ required to indent a bonded layer on its Poisson’s ratio, calculated for conical (a, b) and paraboloidal (c, d) punches. The values of $\Lambda$ were normalized by $\Lambda^\infty$, which is dimensionless load calculated under the assumption that the sample thickness does not influence the parameters of contact. For a conical punch, $\Lambda^\infty = \frac{2}{\pi} \kappa^2$ (Sneddon’s model, eq. (33)), for paraboloidal $\Lambda^\infty = \frac{4}{3} \kappa^2$ (Hertz’s model, eq. (35)).

Formulae for load suitable for layers of arbitrary thickness mitigate problems that arise during implementation of popular procedures for analysis of force-distance curves. To calculate Young’s modulus, it is necessary to identify the point of initial contact $(z_0, P_0)$ between the tip and the sample. A force-distance curve consists of the off-contact ($z < z_0$) and in-contact ($z \geq z_0$) regions. Using standard procedures of the contact point identification, it is necessary to assume a theoretical model of the load-indentation relation suitable for a wide range of values of $\tau$, even for $\tau$ larger than its maximal value reached during the experiment. In the methods based on a grid-search [45], proposed in [46], every point of the curve is treated as a trial contact point. The load-indentation data $\{\delta_i, P_i\}$ are calculated from the corresponding trial in-contact region. Theoretical model of $P(\delta)$ is fitted to $\{\delta_i, P_i\}$ using least squares, while a polynomial is fitted to the corresponding off-contact region. The trail point which gives the lowest total sum of squares of residuals, from both fits, is selected as the final contact point estimate. The trail points located before the actual contact point yield values of $\delta$ and $\tau$ outside their actual range, so that expressions for load valid over wide range of $\tau$ are needed. Formulae for load that combine asymptotic equations and numerical approximations appears to be well suited for this purpose.
The numerical solutions of the problem of a punch indenting a linear-elastic layer have been reported in a few studies, although only for relatively small values of $\tau$. Load required for conical indentation of a layer was calculated numerically in [12] and tabulated for selected values of $\tau \leq 20$. These results are in very good agreement with our numerical computations, with discrepancy below 0.053%. Load necessary for paraboloidal indentation, plotted for $\tau \leq 7$ in [38] also agrees well with our calculations, both for bonded and non-bonded layers. Tabulated values of load required for paraboloidal indentation can be found in [39] for relatively thick ($\tau \leq 3$) bonded layers. The discrepancy between those values and our results is below 0.44%.

We also compared several published approximate formulas for load, derived for the problems specified by the boundary conditions [1] - [7], with the Nyström method solutions of the corresponding integral equations [8]. The asymptotic equations for load required to indent a layer with a conical punch, published in [22], deviate less than 10% from the numerical solution provided that $\kappa < 3.86$ (for a non-bonded) or $\kappa < 0.68$ (for a bonded later). For tips with semi-included angle $\theta < 34.4^\circ$, the value of $\kappa$ is always below 0.68, even if the tip completely penetrates the layer. However, similar equations proposed
in [4] differ significantly from the numerical solutions and the approximate formulae known from the literature, especially in the case of a bonded layer (fig. 5 a). The equations most commonly used for analysis of force - distance curves recorded with paraboloidal or spherical tips were published in [21]. They can be used to correct the substrate effect provided that the layer is relatively thick and the radius of curvature of the tip is small. For an incompressible layer, the relative error of those equations is below 10% when \( \kappa < 0.39 \) (non-bonded, fig. 4 c) or \( \kappa < 0.31 \) (bonded layer, fig. 5 c). The discrepancy between the numerical solution and the equations for load, derived in [20], [38] and [26] for a very thin, non-bonded layer indented with a paraboloid, quickly decreases with \( \kappa \) (fig. 4 d). However, an analogous equation derived in [44] and quoted as eq. (15) in [21] differ from our numerical results by a factor of 1/2. The approximate formulas proposed in [40], [38] and [26] for a bonded, incompressible layer agree with the numerical results, although the decrease of the relative error is slow (fig. 5 d).

Based on the solutions of the Lebedev - Ufliand equation, we can formulate general predictions relevant for analysis of force - distance curve, in particular about potential pitfalls in use of large spherical probes for nearly - incompressible thin samples. The advantages of paraboloidal or spherical AFM tips are well known [47]. Spheres of polysterene or glass can be attached at the free end of a cantilever [48–50]. Such tips are known as colloidal probes. They induce smaller stresses in the material than sharp pyramidal tips. In addition, colloidal probes are axisymmetric, so analysis of their contact with the sample is relatively simple. However, our numerical results indicate that the effect of the rigid substrate is much more pronounced if spherical tips are used instead of conical ones. It is often assumed that the effect of the substrate can be disregarded if the maximal indentation depth is smaller than 10% of sample thickness. For conical punches, the error due to the substrate effect can be regarded as a function of \( \frac{\delta}{h} \) for fixed \( \theta \) (fig. 8 a, b). For paraboloidal punches, the magnitude of the substrate effect can be treated as a function of \( \frac{\delta}{h} \) only when the ratio \( \frac{R}{h} \) is fixed. The influence of substrate may be substantial even if indentations are shallow (fig. 8 c, d). Thus, it is advisable to always take into account the presence of the rigid substrate during analysis of force - distance curves recorded with a paraboloidal or spherical probe. When radius of such a probe is large compared to the layer thickness, calculation of load necessary to indent a nearly incompressible layer (i.e. with \( \nu \) close to 0.5) requires accurate values of Poisson’s ratio of the sample (fig. 7 c, d). For hydrogels ([51–53]) and biological tissues ([54–56]), \( \nu \) is usually between 0.35 - 0.5, often close to 0.5. The accurate value of Poisson’s ratio of the sample is rarely available during analysis of force - distance curves. Thus, it appears advisable to avoid probes of radius much larger than sample thickness in experiments with nearly-incompressible materials. Another solution could be including Poisson’s ratio as an additional fitting parameter during force curve analysis. It is worthwhile to investigate experimentally whether and when such an approach can be used to extract Poisson’s ratio of thin layers from AFM recordings.
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Appendix A. Approximations of the kernel of the Lebedev-Ufliand equation

To estimate the effect of the rigid substrate on load and contact radius, it is necessary to numerically evaluate the kernel $K(x,t;\tau)$ for multiple pairs of $x$ and $t$. $K$ can be expressed using $\Omega(y;\tau)$, as in (10). We will show how to approximate $\Omega$ with a linear combination of the Christov functions.

$\Omega$ is a non-elementary cosine transform of the weight function $\omega$. Approximating $\omega$ with a linear combination of functions that possess elementary cosine transforms, we will obtain an elementary approximation of $\Omega$. We will start with rescaling $\omega(p;\tau)$

$$\tilde{\omega}(x) = \omega(\tau x;\tau) \quad (A.1)$$

In accordance with (12) and (13), $\tau$ in the argument $\tau x$ cancels with the parameter $\tau$. Thus, $\tilde{\omega}(x)$ depends only on $x$. The cosine transform of $\tilde{\omega}$ will be denoted by $\tilde{\Omega}$

$$\tilde{\Omega}(w) = \int_0^\infty \tilde{\omega}(x) \cos(wx) \, dx \quad (A.2)$$

The transforms $\tilde{\Omega}$ and $\Omega$ are related by

$$\tilde{\Omega}(w) = \frac{\Omega(\frac{w}{\tau};\tau)}{\tau} \quad (A.3)$$

As $\tilde{\omega}(x)$ decreases exponentially with $x$, we will approximate it with a combination of exponential functions. Investigating the problem of a thin layer with the Green’s function method, Li i Dempsey [57] proposed approximating $\tilde{\omega}(x)$ as

$$\tilde{\omega}(x) \approx \sum_{n=1}^N a_n e^{-nux} \quad u > 0 \quad (A.4)$$

The coefficients $a_n$ were calculated using the least squares method. The Green’s function was expressed as a Hankel transform of an expression containing $\tilde{\omega}$ as a factor. The approximation (A.4) allowed for calculating the Green’s function as a combination of elliptic integrals.

An approximation of $\Omega$, obtained as a cosine transform of (A.4), contains only elementary functions

$$\Omega(y;\tau) \approx \tau \sum_{n=1}^N a_n \frac{nu}{n^2u^2 + \tau^2y^2} \quad (A.5)$$

The basis of functions used in (A.4) is not orthogonal. To simplify calculation of the expansion coefficients, we will use the orthogonal basis of Laguerre functions.
\( \ell_n(x; u) \), defined as products of normalized Laguerre polynomials \( L_n(ux) \) and exponentials

\[
\ell_n(x; u) = e^{-\frac{ux^2}{2}} L_n(ux) \quad u > 0, \quad n = 0, 1, 2, \ldots
\]  

(A.6)
The basis \( \{L_n(x)\}_{n=0}^\infty \) is orthogonal in \([0, \infty)\). For any non-negative integers \( n \) and \( m \)

\[
\int_0^\infty \ell_n(x; u) \ell_m(x; u) \, dx = \frac{1}{u} \delta_{nm}
\]

(A.7)
where \( \delta_{nm} \) is the Kronecker delta. The expansion of \( \tilde{\omega}(x) \) with respect to the basis \( \{\ell_n(x; u)\}_{n=0}^\infty \), truncated to the first \( N+1 \) terms, can be written as

\[
\tilde{\omega}(x) \approx u \sum_{n=0}^N \int_0^\infty \tilde{\omega}(s) \ell_n(s; u) \, ds \ell_n(x; u) \quad u > 0
\]

(A.8)
Substituting \( \omega \) for \( \tilde{\omega} \) on the left hand side of (A.8), we obtain

\[
\omega(p; \tau) \approx u \sum_{n=0}^N \int_0^\infty \tilde{\omega}(s) \ell_n(s; u) \, ds \ell_n(s; \frac{p}{\tau}; u)
\]

(A.9)
Cosine transforms of \( \ell_n \) can be expressed as [58]

\[
\int_0^\infty \ell_n\left(\frac{p}{\tau}; u\right) \cos(py) \, dp = \frac{\tau \frac{u}{2}}{(\frac{y}{\tau})^2 + (y\tau)^2} U_{2n}\left(\frac{y\tau}{\sqrt{(\frac{y}{\tau})^2 + (y\tau)^2}}\right) = \frac{2\tau}{u} CC_{2n}\left(y\tau; \frac{u}{2}\right)
\]

(A.10)
(A.11)
where \( U_{2n} \) is a Chebyshev polynomial of the second kind of the \( 2n \)-th degree. In Chebyshev polynomials of even degree all odd coefficients are equal to zero. Thus, the expression on the right hand side of (A.10) is a rational function. \( CC_{2n} \) denotes an even Christov function [59, 60]. It can be defined using Chebyshev polynomials of the second kind

\[
CC_{2n}(x; L) = \frac{L^2}{L^2 + x^2} U_{2n}\left(\frac{x}{\sqrt{L^2 + x^2}}\right)
\]

(A.12)
where \( n \) is a non-negative integer and \( L \) is a positive real number. Combining (11), (A.9) and (A.11), we obtain

\[
\Omega(y; \tau) \approx 2\tau \sum_{n=0}^N \left[ \int_0^\infty \tilde{\omega}(s) \ell_n(s; u) \, ds \right] CC_{2n}\left(y\tau; \frac{u}{2}\right)
\]

(A.13)

It can be shown that the approximation (A.13) is equal to the truncated expansion of \( \Omega \) with respect to the basis of the even Christov functions \( \{CC_{2n}\}_{n=0}^\infty \). This basis is orthogonal in \((-\infty, \infty)\) with weight equal to 1 [59]. In accordance
with \( \tilde{\Omega} \). \( \Omega \) depends on \( \tau \) only through \( \omega(p; \tau) \), which in turn depends on \( \tilde{\omega} \).

After change of variables, \( \Omega \) can be regarded as a function of \( w = y\tau \), denoted as \( \tilde{\Omega} \) and given by \( \tilde{\Omega}(y) \) in \((-2, 2)\). To approximate \( \tilde{\Omega}(w) \) in \((-\infty, \infty)\). As \( \tilde{\Omega} \) is an even function, it can be expanded with respect to \( \{ CC_{2n}(w; \frac{u}{2}) \}_{n=0}^{\infty} \)

\[
\tilde{\Omega}(w) \approx \sum_{n=0}^{N} \left[ \int_{-\infty}^{\infty} \frac{4}{\pi u} \tilde{\Omega}(t) CC_{2n}(t; \frac{u}{2}) \, dt \right] CC_{2n}(w; \frac{u}{2})
\]  

(A.14)

We will express (A.14) in a form similar to that of (A.13). \( \tilde{\Omega}(t) \) and \( CC_{2n}(t; L) \) are even, so the integrand in (A.14) is even. The integral in \((-\infty, \infty)\) can be replaced by an integral in \((0, \infty)\). We will also replace \( \tilde{\Omega} \) with \( \Omega \), in accordance with (A.3). After simplification

\[
\Omega(y; \tau) \approx 2\tau \sum_{n=0}^{N} \left[ \int_{0}^{\infty} \frac{4}{\pi u} \tilde{\Omega}(t) CC_{2n}(t; \frac{u}{2}) \, dt \right] CC_{2n}(y\tau; \frac{u}{2})
\]  

(A.15)

In accordance with Parseval’s theorem for cosine transforms, for any smooth functions \( g \) and \( h \), which are integrable and square-integrable, it holds that

\[
\int_{0}^{\infty} g(s)h(s) \, ds = \frac{2}{\pi} \int_{0}^{\infty} \left( \int_{0}^{\infty} g(s) \cos(st) \, ds \right) \left( \int_{0}^{\infty} h(s) \cos(st) \, ds \right) \, dt
\]  

(A.16)

\( \tilde{\Omega} \) is a cosine transform of \( \tilde{\omega} \), while \( \frac{2}{\pi} CC_{2n}(t; \frac{u}{2}) \) is a cosine transform of \( \ell_n(s; u) \).

Combining (A.2) and (A.11) with Parseval’s theorem, we obtain

\[
\int_{0}^{\infty} \tilde{\omega}(s)\ell_n(s; u) \, ds = \frac{4}{\pi u} \int_{0}^{\infty} \tilde{\Omega}(t) CC_{2n}(t; \frac{u}{2}) \, dt
\]  

(A.17)
Hence the direct expansion (A.15) of $\Omega$ with respect to the basis of the even Christov functions is equal to (A.13). However, calculating the expansion coefficients from (A.13) is much faster. In (A.13), the integrand is an elementary function $\tilde{\omega}$, while in (A.15), the integrand is itself a non-elementary integral.

The relative error $\epsilon$ of an approximation $\tilde{\Omega}_N$ was calculated as $\epsilon = \frac{\|\tilde{\Omega} - \tilde{\Omega}_N\|_2}{\|\tilde{\Omega}\|_2}$, where $\|\cdot\|_2$ is the $L^2$ norm. Numerical results indicate that the relative error of the expansion of $\tilde{\Omega}$ into the even Christov functions decreases exponentially with the number of terms (Fig. A.9). To illustrate the importance of choice of basis functions for approximation of $\tilde{\Omega}$, we compared the approximations employing the Christov functions with the expansion in series of the Hermite functions, which are also orthogonal in $(-\infty, \infty)$. The relative error of an expansion into the Hermite functions is larger for any $N$ tested.
Appendix B. Supporting figure

Figure S1: The influence of the number of nodes $N$ in the Gauss - Chebyshev - Radau quadrature on the equilibrium value of $\tau$, calculated from the Nyström method solutions $\chi$ of the Lebedev - Ufliand equation. The equilibrium value of $\tau$ was found as a root of $\chi(1; \tau)$, using Newton's algorithm. The plotted values are $\epsilon = (\tau_N - \tau_{1600})/\tau_{1600}$, where $\tau_N$ is the estimation obtained with $N$ nodes. Calculations were performed for a paraboloidal tip indenting a non-bonded (results for different values of reduced indentation depth $\kappa$ are given in a, c, e) or bonded incompressible (b, d, f) layer.
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